Applicant : Steven Reppert et al. Attorney's Docket No.: 10217-250003 MGH-0823.3: BMS X22c

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* againments to the Claims:

This listing of claims replaces all prior versions and listings of claims in the application:

<u>Listing of Claims</u>:

1-52. (cancelled)

53. (currently amended) A method of testing a candidate compound for the ability to act as an agonist of a high-affinity melatonin receptor ligand, said method comprising:

a) contacting said candidate compound with providing a cell comprising an expression vector encoding a high affinity melatonin receptor protein comprising an unino acid sequence substantially identical at least 80% identical to that of SEQ ID NO:12, or a melatonin binding fragment thereof, wherein the cell expresses on its surface said high affinity melatonin receptor protein or melatonin binding fragment thereof:

b) contacting said melatonin receptor protein or melatonin binding fragment with the candidate compound;

b) c) measuring intracellular cAMP concentration in said cell; and

e) d) where said contacting causes a decrease in intracellular cAMP concentration, identifying said candidate compound as an agonist of a high-affinity melatonin receptor ligand

54-77. (cancelled)

78. (currently amended) A method of testing a candidate compound for the

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ability to act as an agonist of a high affinity melatonin receptor ligand, said method comprising:

a) contacting said candidate compound with providing a cell comprising an expression vector encoding a high-affinity melatonin receptor protein comprising an amino acid sequence substantially identical at least 80% identical to that of SEQ ID 50.6, or a melatonin binding fragment thereof, wherein the cell expresses on its surface said high-affinity melatonin receptor protein or melatonin binding fragment thereof;

b) contacting said melatonin receptor protein or melatonin binding fragment with the candidate compound:

b) c) measuring intracellular cAMP concentration in said cell; and

e) d) where said contacting causes a decrease in intracellular cAMP concentration, identifying said candidate compound as an agonist of a high affinity melatonin receptor ligand.

79. (cancelled)

80. (currently amended) A method of testing a candidate compound for the ability to act as an agonist of a high affinity melatonin receptor ligand, said method comprising:

a) contacting said candidate compound with providing a cell comprising an expression vector encoding a high affinity melatonin receptor protein, or a melatonin binding fragment thereof, wherein the expression vector comprises a sequence that hybridizes to a probe having the sequence of the complement of SEQ ID NO:5 under the following conditions: hybridization for 50% formamide. 1 M sodium chloride, 1% SDS, 10% dextran sulfate, 100 µg/ml denatured salmon sperm at 42 °C, and filters washed in 2x SSC, 1% SDS at 65 °C for 1 hour, and wherein the cell expresses on its surface said melatonin receptor protein or melatonin binding fragment;

b) contacting said melatonin receptor protein or melatonin binding fragment with the candidate compound;

b) c) measuring intracellular cAMP concentration in said cell; and

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e) d) where said contacting causes a decrease in intracellular cAMP concentration. identifying said candidate compound as an agonist of a high affinity melatonin receptor ligand.

- 81. (currently amended) A method of testing a candidate compound for the ability to act an agonist of a high affinity melatonin receptor ligand, said method comprising:
- a) contacting said candidate compound with providing a cell comprising an expression vector encoding a high affinity melatonin receptor protein, or a melatonin binding fragment thereof, wherein the expression vector comprises a sequence that hybridizes to a probe having the sequence of the complement of SEQ ID NO:5 under the following conditions: hybridization in 25% formamide, 1 M sodium chloride, 1% SDS, 10% dextran sulfate, 100 µg/ml denatured salmon sperm at 42 °C, and filters washed in 2x SSC, 1% SDS at 55 °C for 1 hour, and wherein the cell expresses on its surface said melatonin receptor protein or melatonin binding fragment:

b) contacting said melatonin receptor protein or melatonin binding fragment with the candidate compound;

- b) c) measuring intracellular cAMP concentration in said cell; and
- \leftrightarrow <u>d)</u> where said contacting causes a decrease in intracellular cAMP concentration, identifying said candidate compound as an agonist of a high affinity melatonin receptor ligand.
- 82. (previously presented) The method of claim 81, wherein the expression vector comprises the sequence of SEQ ID NO:5.
- 83. (currently amended) A method of testing a candidate compound for the ability to act as an agonist of a high affinity melatonin receptor ligand, said method comprising:
- a) contacting said candidate compound with providing a cell comprising an expression vector encoding a high affinity melatonin receptor protein, or a melatonin binding fragment thereof, wherein the expression vector comprises a sequence that hybridizes to a probe having the sequence of the complement of SEQ ID NO:11 under the following conditions: hybridization in 50% formamide, 1 M sodium chloride, 1% SDS, 10% dextran sulfate, 100 µg/ml

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denatured salmon sperm at 42 °C, and filters washed in 2x SSC, 1% SDS at 65 °C for 1 hour, and wherein the cell expresses on its surface said melatonin receptor protein or melatonin building fragment;

b) contacting said melatonin receptor protein or melatonin binding fragment with the candidate compound:

- b) c) measuring intracellular cAMP concentration in said cell; and
- e) d) where said contacting causes a decrease in intracellular cAMP concentration, identifying said candidate compound as an agonist of a high affinity melatonin receptor ligand.
- \$4. (currently amended) A method of testing a candidate compound for the ability to act as an agonist of a high affinity melatonin receptor ligand, said method comprising:
- a) contacting said candidate compound with providing a cell comprising an expression vector encoding a high-affinity melatonin receptor protein. or a melatonin binding fragment thereof, wherein the expression vector comprises a sequence that hybridizes to a probe having the sequence of the complement of SEQ ID NO:11 under the following conditions: hybridization in 25% formamide, 1 M sodium chloride, 1% SDS, 10% dextran sulfate, 100 µg/ml denatured salmon sperm at 42 °C, and filters washed in 2x SSC, 1% SDS at 55 °C for 1 hour, and wherein the cell expresses on its surface said melatonin receptor protein or melatonin binding fragment;

b) contacting said melatonin receptor protein or melatonin binding fragment with the candidate compound;

- b) c) measuring intracellular cAMP concentration in said cell; and
- \leftrightarrow <u>d)</u> where said contacting causes a decrease in intracellular cAMP concentration, identifying said candidate compound as an agonist of a high affinity melatonin receptor ligand. --
- 85. (previously presented) The method of claim 84, wherein the expression vector comprises the sequence of SEQ ID NO:11.

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86. (currently amended) A method of testing a candidate compound for the ability to act as an agonist of a high affinity melatonin receptor ligand, said method comprising:

a) contacting said candidate compound with providing a cell comprising an expression contaction choosing a high-affinity melatonin receptor protein that consists of the amino acid sequence of SEQ ID NO:12, or a melatonin binding fragment thereof, wherein the cell expresses on its surface said high-affinity melatonin receptor protein or melatonin binding fragment thereof:

b) contacting said melatonin receptor protein or melatonin binding fragment with the candidate compound;

- b) c) measuring intracellular cAMP concentration in said cell; and
- e) d) where said contacting causes a decrease in intracellular cAMP concentration, identifying said candidate compound as an agonist of a high affinity melatonin receptor ligand.
- 87. (currently amended) A method of testing a candidate compound for the ability to act as an agonist of a high affinity melatonin receptor ligand, said method comprising:
- a) contacting said candidate compound with providing a cell comprising an expression vector encoding a high-affinity melatonin receptor protein comprising the amino acid sequence of SEQ ID NO:6, or a melatonin binding fragment thereof, wherein the cell expresses on its surface said high affinity melatonin receptor protein or melatonin binding fragment thereof;

b) contacting said melatonin receptor protein or melatonin binding fragment with the candidate compound;

- b) c) measuring intracellular cAMP concentration in said cell; and
- e) d) where said contacting causes a decrease in intracellular cAMP concentration, identifying said candidate compound as an agonist of a high affinity melatonin receptor ligand.
- 88. (currently amended) A method of testing a candidate compound for the ability to act as an agonist of a high affinity melatonin receptor ligand, said method comprising:

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a) contacting said candidate compound with providing a cell comprising an expression vector encoding a high affinity melatonin receptor protein that comprises the amino acid sequence of SEQ ID NO:12, or a melatonin binding fragment thereof, wherein the cell expresses on its surface said high affinity melatonin receptor protein or melatonin binding fragment diereof)

b) contacting said melatonin receptor protein or melatonin binding fragment with the candidate compound;

- b) c) measuring intracellular cAMP concentration in said cell; and
- e) d) where said contacting causes a decrease in intracellular cAMP concentration, identifying said candidate compound as an agonist of a high affinity melatonin receptor ligand.
- 89 (currently amended). A method of testing a candidate compound for the ability to act as an agonist of a high affinity melatonin receptor ligand, said method comprising:
- a) contacting said candidate compound with providing a cell comprising an expression vector encoding a high affinity melatonin receptor protein consisting of the amino acid sequence of SEQ ID NO:6, or a melatonin binding fragment thereof, wherein the cell expresses on its surface said high affinity melatonin receptor protein or melatonin binding fragment thereof;
- b) contacting said melatonin receptor protein or melatonin binding fragment with the candidate compound;
 - b) c) measuring intracellular cAMP concentration in said cell; and
- e) d) where said contacting causes a decrease in intracellular cAMP concentration, identifying said candidate compound as an agonist of a high affinity melatonin receptor ligand.
- 90. (new) The method of claim 53, wherein the melatonin receptor protein differs from SEQ ID NO:12 only by conservative substitutions.
- 91. (new) The method of claim 78, wherein the melatonin receptor protein differs from SEQ ID NO:6 only by conservative substitutions.